Effects of Inadequate Amino Acid Mixture Intake on Nutrient Supply of Adult Patients with Phenylketonuria

Hochuli, Michel ; Bollhalder, Sandra ; Thierer, Carina ; Refardt, Julie ; Gerber, Philipp ; Baumgartner, Matthias R

Abstract: BACKGROUND Adult phenylketonuria (PKU) patients often reduce their intake of amino acid mixture (AAM) to less than the prescribed amounts. Effects of reduced AAM intake on nutrient supply were evaluated. METHODS Nutrient intake was calculated in 20 adult PKU patients based on a structured food record and complemented by laboratory assessment of nutritional status. Patients were classified into 2 groups, (A) regular AAM intake, or (B) AAM intake below calculated requirements. RESULTS Group B consumed a higher proportion of natural protein (60 ± 23 vs. 33 ± 12%, p = 0.002); however, the total protein intake was below the recommended amounts in 60% of patients in group B versus 7% in group A (p = 0.03). Fat intake was higher in group B (39 ± 9% of energy vs. 31 ± 6%, p = 0.03), mainly from saturated fats. Selenium, folate, and vitamin B12 intake was below the recommended intake in group B. However, serum concentrations of these analytes remained within the normal range in both groups, although vitamin B12 levels were lower in group B. Plasma tyrosine correlated with AAM intake, and hydroxyproline correlated with the amount of natural protein consumed. CONCLUSION Relaxed AAM intake resulted in insufficient nutrient supply, despite a compensatory increase in consumption of natural protein. Care needs to be taken to ensure adequate nutrition in adults with PKU.

DOI: https://doi.org/10.1159/000479746

Original Paper

Annals of Nutrition & Metabolism

Ann Nutr Metab 2017;71:129–135
DOI: 10.1159/000479746

Received: February 2, 2017
Accepted after revision: July 24, 2017
Published online: August 26, 2017

Effects of Inadequate Amino Acid Mixture Intake on Nutrient Supply of Adult Patients with Phenylketonuria

Michel Hochuli[a, c] Sandra Bollhalder[a] Carina Thierer[a] Julie Refardt[d] Philipp Gerber[a] Matthias R. Baumgartner[b, c]

[a]Division of Endocrinology, Diabetes, and Clinical Nutrition, University Hospital Zurich, Zurich, [b]Division of Metabolism and Children's Research Center, University Children's Hospital, Zurich, and [c]Radiz – Rare Disease Initiative Zurich, Clinical Research Priority Program for Rare Diseases, University of Zurich, Zurich, and [d]Clinic of Endocrinology, Diabetes and Metabolism, University Hospital Basel, Basel, Switzerland

Keywords
Phenylketonuria · Adult · Nutrition · Nutrient · Vitamin · Treatment · Amino acids · Supplement · Compliance · Hydroxyproline

Abstract
Background: Adult phenylketonuria (PKU) patients often reduce their intake of amino acid mixture (AAM) to less than the prescribed amounts. Effects of reduced AAM intake on nutrient supply were evaluated. ***Methods:*** Nutrient intake was calculated in 20 adult PKU patients based on a structured food record and complemented by laboratory assessment of nutritional status. Patients were classified into 2 groups, (A) regular AAM intake, or (B) AAM intake below calculated requirements. ***Results:*** Group B consumed a higher proportion of natural protein (60 ± 23 vs. 33 ± 12%, p = 0.002); however, the total protein intake was below the recommended amounts in 60% of patients in group B versus 7% in group A (p = 0.03). Fat intake was higher in group B (39 ± 9% of energy vs. 31 ± 6%, p = 0.03), mainly from saturated fats. Selenium, folate, and vitamin B12 intake was below the recommended intake in group B. However, serum concentrations of these analytes remained within the normal range in both groups, although vitamin B12 levels were lower in group B. Plasma tyrosine correlated with AAM intake, and hydroxyproline correlated with the amount of natural protein consumed. ***Conclusion:*** Relaxed AAM intake resulted in insufficient nutrient supply, despite a compensatory increase in consumption of natural protein. Care needs to be taken to ensure adequate nutrition in adults with PKU.

Introduction

Phenylketonuria (PKU, OMIM 261600) is an inherited disorder of phenylalanine metabolism. PKU results from a genetic defect of the enzyme phenylalanine hydroxylase, which converts phenylalanine (Phe) to tyrosine as the first step of the Phe degradation pathway [1, 2]. Newborn screening for PKU with the early introduction of dietary therapy has been a great success, preventing the severe neurological damage that occurs in children with high Phe levels when PKU is left untreated. In dietary treatment of PKU, natural protein intake is restricted (low-Phe diet), and a Phe-free amino acid mixture (AAM) is supplemented to ensure adequate protein supply [3]. Trace elements, minerals, and vitamins are added to the AAM to compensate for nutritional imbal-

E-Mail karger@karger.com
www.karger.com/anm

Michel Hochuli, MD, PhD
Division of Endocrinology, Diabetes, and Clinical Nutrition
University Hospital Zurich
Rämistrasse 100, CH–8091 Zurich (Switzerland)
E-Mail michel.hochuli@usz.ch

ances resulting from dietary restrictions [4]. The amount of natural protein allowed in the diet depends on the residual enzyme activity and the defined plasma Phe concentration target. Strict control of Phe levels is mandatory in childhood and early adolescence to ensure normal development and to avoid neurological damage, as well as in pregnancy to prevent Phe fetopathy. For adults, current guidelines recommend life-long dietary treatment [5, 6]. National recommendations regarding the upper target blood Phe concentration for adults outside pregnancy are somewhat heterogeneous. The advised upper limit in Switzerland is 600 μmol/L, which is in accordance with the recently published European guideline [6]. However, many patients with good Phe control in childhood and adolescence will have difficulties to maintain a rigid diet in adulthood [7–10]. Adults often find it difficult to integrate a strict diet, including regular AAM intake, into social and professional life. They frequently relax their protein-restricted diet and reduce AAM intake below prescribed amounts. Commonly, patients maintain their habitual dietary practices with a variable degree of natural protein restriction despite relaxed AAM intake, which constitutes a risk for nutritional imbalances and deficiencies [11, 12]. In the present prospective cross-sectional study, eating habits, nutritional status, and selected nutritional biomarkers were evaluated in a cohort of adult PKU patients, in relation to their treatment compliance regarding AAM intake.

Materials and Methods

Study Design

This study was planned in a prospective cross-sectional design. Patients with classical or moderate PKU were recruited during a regular consultation in the outpatient clinics of the University Hospitals of Zurich and Basel. Study participants were asked to fill in a questionnaire about current modalities and frequency of AAM intake, and were asked to provide a detailed 4-day food record within 4 weeks after the consultation when recruited for the study. Anthropometric measurements and laboratory assessment were done as part of standard medical care on the occasion of this consultation. Laboratory testing included biochemical parameters, such as creatinine, C-reactive protein, albumine, prealbumine, selected minerals and trace elements (ferritin, selenium, zinc), vitamins (vitamin B12, folic acid, 25-OH vitamin D), as well as a complete plasma amino acid profile. Amino acid profiles were measured from heparin plasma by ion-exchange high-performance liquid chromatography. Venous blood samples were collected in the non-fasting state prior to the next main meal (mainly before lunch), 3–4 h after the last AAM intake, according to the typical clinical routine in the outpatient department. All procedures followed were in accordance with the ethical standards of the responsible committee on human experimentation (registration number KEK ZH 2013-0120) and with the Helsinki Declaration of 1975 as revised in 2000. The study was registered at the database ClinicalTrials.gov (NCT01879995).

Patients and Methods/Study Groups

Male or female patients with classical or moderate PKU aged >18 years and willing to provide a detailed food record were eligible for the study. None of the patients was on a treatment with sapropterin (BH4). Twenty-four patients (17 classical, 7 moderate PKU, 14 male, 10 female) were recruited, 21 patients from the outpatient clinic in Zurich, and 3 from Basel. All patients participating in the study returned the food records and questionnaires. Patients were classified into 2 groups, (A) regular AAM intake (i.e., total daily amount of AAM according to prescription, taken in 2 or 3 portions per day) and (B) AAM intake below recommendations. This classification was based on the patient interview at the time of recruitment (when blood sampling was done), as well as on the information obtained from the individual study questionnaires filled in after this consultation. Answers in the questionnaires were checked for consistency with the patient interview and medical history. Patients were instructed not to change dietary habits between the consultation and the time when food intake was recorded. Four patients with moderate PKU had a Phe tolerance high enough to reach blood Phe levels <600 μmol/L without requiring any AAM to cover their protein needs. These 4 patients were omitted from these 2 groups, but the data were used for the correlation of natural protein intake with plasma hydroxyproline concentrations. Informed consent was obtained from all patients for being included in the study. The first patient was recruited in June 2013 and the last in February 2015.

Assessment of Nutrient Intake

Nutrient intake was calculated based on a detailed 4-day diet record including the type and quantity of all foods, beverages, and the AAM taken within 4 consecutive days. Whenever possible, the quantity of food was weighed by the patients, using a digital kitchen scale. One of the 4 protocolled days was required to be a weekend day, and the other 3 days weekdays. Patients were asked to send in a dried blood card for measurement of Phe after completion of the food record. Macro- and micronutrient intake was calculated based on the food records, using the Food Control Management System "DIÄT-2000" software (Soft&Hard, D. Beyer) [12]. Reference values for nutrient intake were based on the 2015 D-A-CH recommendations [13]. Recommended protein intake, including AAM, was calculated according to the D-A-CH reference value for adults (0.8 g/kg body weight), with an empirical increment of +10%. This empirical increment corresponded to the usual practice of establishing dietary treatment plans in the participating clinics at the time when this study was conducted. Patients were allowed animal protein according to their individual Phe tolerance and as long as Phe values were well controlled.

Statistics

Data are presented as arithmetic means with SD or ranges. Statistical analysis was performed using the statistical package SPSS 22 (IBM SPSS Statistics). A 2-sided value of $p < 0.05$ was considered significant. For comparison of continuous variables between groups, an independent samples *t*-test or one-way analysis of variance was applied. Phe values of individual patients were compared using paired *t*-tests. Non-normally distributed data were log-

transformed prior to statistical testing. Fisher's exact test was used to compare categorical variables presented as percentages. One-way analysis of variance with linear contrast was used to test for a linear trend of hydroxyproline or tyrosine concentrations across categories of natural protein or AAM intake. Post-hoc Bonferroni correction was applied to account for multiple comparisons.

Results

The present analysis is based on a cohort of 20 PKU patients, 15 were allocated to group A (regular AAM intake, i.e., a total daily amount of AAM according to prescription, taken in 3 or fewer of portions per day) and 5 to group B (AAM intake below the prescribed amount). The mean age was 32 ± 12 years in group A (median 31 years, range 18–64 years) and 39 ± 8.4 years in group B (median 39 years, range 30–50 years; ns). Group B had a higher proportion of males (47% group A, 80% group B), albeit not statistically significant. Body mass index was significantly lower in group B with irregular AAM intake (body mass index: group A 24.6 ± 4.3 kg/m^2, group B 20.6 ± 2.1 kg/m^2, $p = 0.04$), although total energy intake during the protocolled period was similar in both groups (see below). The plasma Phe concentration at the time of the consultation (measured as part of the complete plasma amino acid profile by ion-exchange high-performance liquid chromatography) was somewhat above the recommended upper target in Switzerland of 600 µmol/L, with a statistically nonsignificant trend toward higher Phe values in the group with irregular AAM intake (group A 650 ± 283, group B 760 ± 350 µmol/L; ns). Plasma Phe values of individual patients were similar to the values obtained in the previous and subsequent consultations. Phe values measured from dried blood spots after the completion of the food records (where available) were not significantly different from the Phe values measured from dried blood spots sent in by patients during the year before they were recruited for the study. The median time interval between the consultation when laboratory assessment was performed and keeping the food record was 8.5 days. Reported Phe tolerance based on pediatric medical charts was similar in both groups (group A 493 ± 183 mg, group B 450 ± 218 mg; ns). However, this value was not available for all study participants.

Nutrient Intake and Laboratory Assessment

The prescribed daily amount of protein from AAM was 0.73 ± 0.2 g/kg body weight for group A and 0.68 ± 0.2 g/kg for group B. The effective mean daily AAM intake was equal to the prescribed amount in group A, whereas AAM intake in group B corresponded to approximately half the prescribed amount (A 0.73 ± 0.1 g/kg vs. B 0.38 ± 0.3 g/kg protein from AAM, $p = 0.001$; Fig. 1). The prescribed AAM were balanced medical PKU products commercially available in Switzerland, mainly concentrated amino acid powders (P-AM 3 [Nutricia GmbH], 14 patients. Milupa PKU 3 [Nutricia GmbH], 3 patients. Milupa PKU 3-advanta [Nutricia GmbH], 2 patients. XPhe smart A [metaX], 1 patient). A few patients replaced single portions of the amino acid powder by portioned or ready-to-drink AAM products (P-AM Anamix [Nutricia GmbH], 1 patient. PKU Lophlex LQ 20 [Nutricia GmbH], 1 patient). There were no relevant differences between the 2 groups with respect to the type and composition of the prescribed AAMs. The daily amount of AAM was taken on average in 2.6 portions per day (range 2.0–3.0) in group A vs. 1.4 portions per day (range 0–2.0) in group B. Although the consumption of natural protein (percent of total protein intake) was higher in group B with irregular AAM intake (60 ± 23 vs. 33 ± 12%, $p = 0.002$), the total protein intake was below the recommended intake in 60% of patients in group B versus 7% in group A ($p = 0.03$). The mean deviation of protein intake from the reference value was + 31.1 ± 27% in group A vs. –0.72 ± 35% in group B ($p = 0.02$). Daily energy intake was not significantly different between both groups (group A: 2167 kcal/day, 32 kcal/kg/day. Group B 2272 kcal/day, 38 kcal/kg/day; ns). However, patients with irregular AAM intake consumed a higher proportion of fat (39 ± 9 vs. 31 ± 6% of total energy intake, $p = 0.03$), mainly from saturated fats (intake 33.3 ± 13 vs. 20.9 ± 7.8 g, $p = 0.017$). The intake of monounsaturated and essential fatty acids was not significantly different between groups. Carbohydrate intake was equal in both groups (52% of total energy).

Supply of calcium, vitamin B12, folate, selenium, and zinc was significantly lower in the group with irregular AAM intake, but only selenium, folate, and vitamin B12 intake was clearly below the recommended intake quantity (Fig. 2). However, despite lower intake, mean serum concentrations of these analytes remained within the normal range in both groups, although vitamin B12 levels were significantly lower in group B but still within normal limits (Table 1). All plasma amino acid concentrations remained within the normal range in both groups, although many patients in group B had a protein intake below the recommended intake (Table 2). There was, however, a trend toward lower concentrations of branched chain amino acids in group B (not significant). Glutamine was significantly lower in group A. Tyrosine (i.e., the product of the deficient enzyme) was lower in group B.

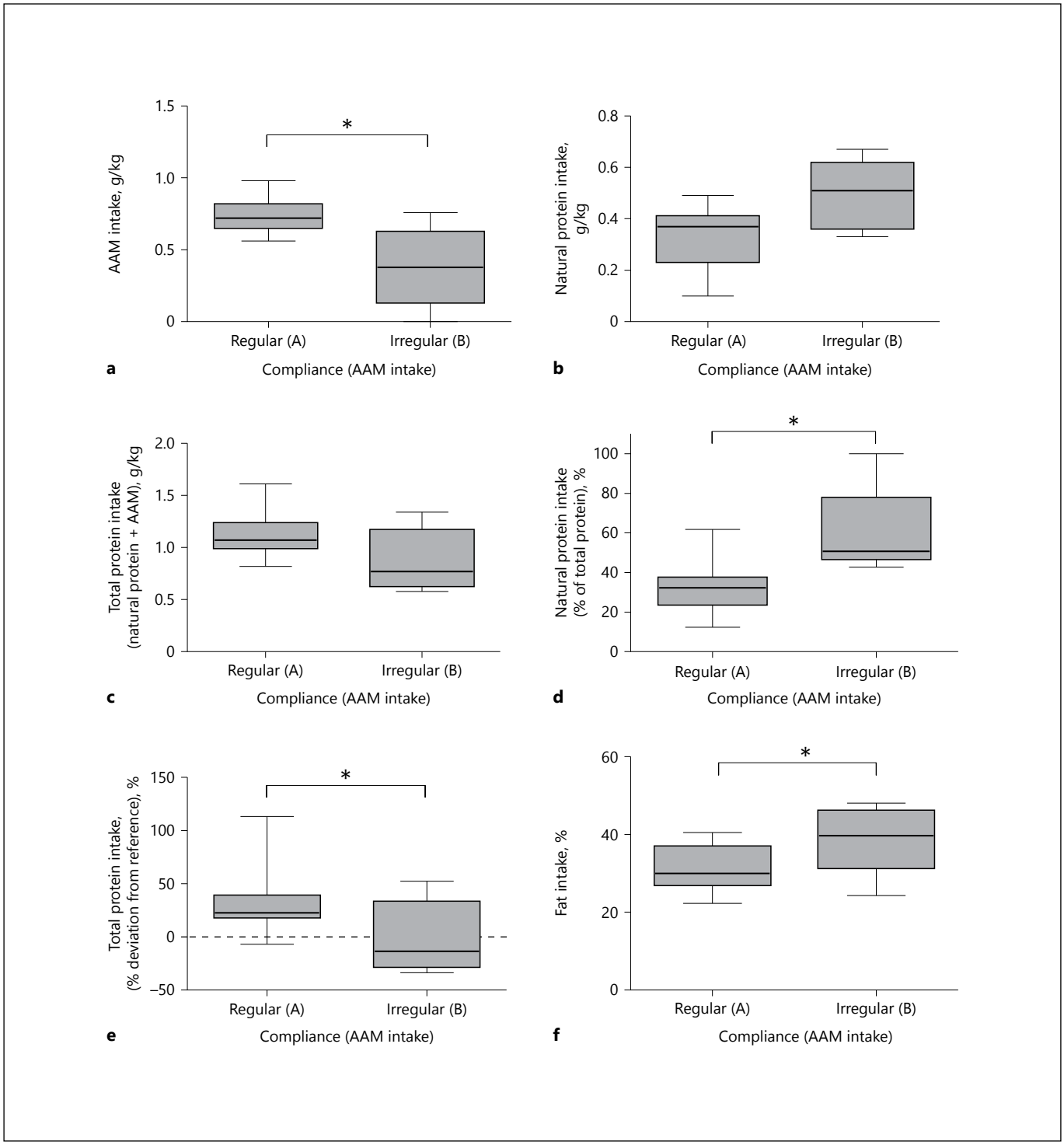

Fig. 1. Macronutrient intake. Protein intake from amino acid mixture (AAM; **a**), natural protein intake (absolute amount, and in percent of total protein intake; **b**, **d**), total protein intake (from AAM and natural protein, absolute amount and as percent deviation from the recommended intake; **c**, **e**), according to treatment compliance regarding AAM intake. The mean daily AAM intake of group B corresponds to approximately half the amount of group A. Despite a compensatory increase in consumption of natural protein, the majority (60%) of patients in group B had a total protein intake below the recommended intake. Fat intake was higher in group B, mainly from saturated (animal) fats (**f**). * Significantly different ($p < 0.05$). Box and whiskers plots with bars representing minimum to maximum values.

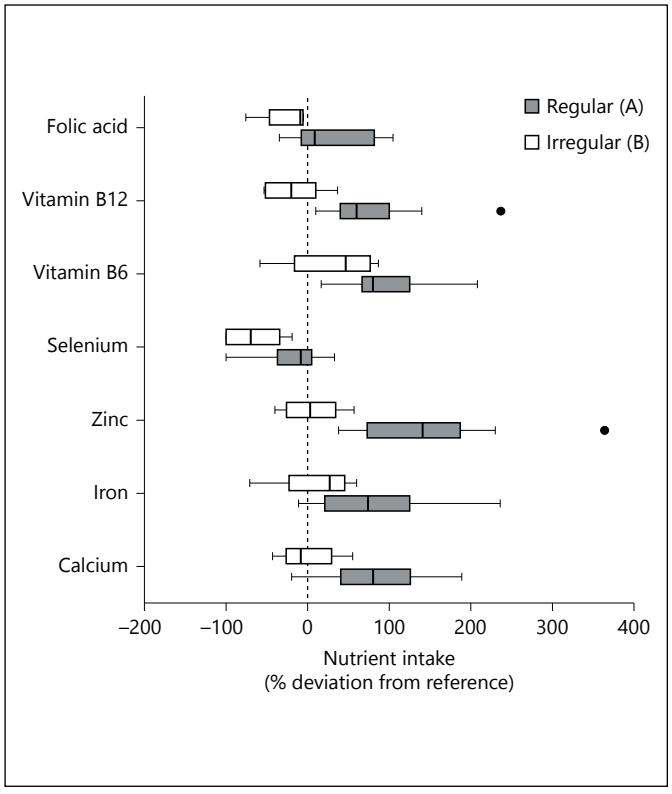

Fig. 2. Intake of selected minerals, trace elements, and vitamins. Supply of calcium, vitamin B12, folate, selenium, and zinc was significantly lower in the group with irregular amino acid mixture intake, but only selenium, folate, and vitamin B12 intake was clearly below the recommended intake. Nutrient intake is presented as percent deviation from the recommended intake. Tukey box and whiskers plot. * Significantly different ($p < 0.05$).

Plasma tyrosine concentrations related to the amount of AAM consumed (Fig. 3). There was a clear association of plasma hydroxyproline concentrations with the proportion of natural protein in the diet (Fig. 3).

Discussion

This study shows that adult PKU patients who relax their intake of AAM have a distinct dietary pattern, with an increased intake of saturated fats and insufficient supply of protein, vitamin B12, and selenium compared to patients with regular AAM intake. Patients with lower AAM intake consumed more natural protein, but this did not fully compensate for the lower supply from the AAM. The higher fat consumption (mainly saturated fats) in the group with irregular AAM intake may be attributed to a higher consumption of animal protein (meat and dairy products).

Table 1. Selected laboratory parameters

	Group (AAM intake)		Normal range
	regular (A)	irregular (B)	
Folic acid	98±290	14±3	>4 µg/L
Vitamin B12	540±208‡	251±75‡	180–914 ng/L
Selenium	0.79±0.20	0.94±0.40	0.8–1.1 µmol/L
Zinc	11.2±2.2	11.8±1.5	9–21 µmol/L
Ferritin	78±76	92±52	15–400 µg/L
25-OH vitamin D	34±10	31±12	>20 µg/L
Albumin	45±6	49	40–49 g/L
Prealbumine	295±40	345±74	200–400 mg/L
Creatinine	71±12	82±25	44–106 µmol/L
C-reactive protein	1.9±2	0.3±0.4	<5 mg/L

Values are given as mean ± SD.
‡ Significantly different between groups.

Table 2. Plasma amino acid concentrations

	Group (AAM intake)		Normal range, µmol/L
	regular (A)	irregular (B)	
Alanine	389±61	418±149	192–508
Arginine	48±25	46±26	45–125
Aspartic acid	10±7	8±5	4–28
Asparagine	38±13	44±7	32–64
Glutamic acid	63±37	64±50	11–59
Glutamine	479±75‡	593±58‡	396–740
Glycine	233±50	341±140	148–324
Histidine	77±13	75±4	58–106
Isoleucine	58±20	44±12	38–94
Leucine	101±32	82±19	76–168
Lysine	165±34	136±23	105–221
Methionine	19±5	17±4	16–36
Phenylalanine	649±283	760±352	38–78
Proline	181±42	201±52	75–307
Serine	94±15	111±21	75–175
Threonine	127±29	104±27	72–192
Tryptophane	36±8	29±4	27–75
Tyrosine	72±24‡‡	42±17‡	40–92
Valine	274±68	221±50	142–278

Values are given as means ± SD.
‡ Significantly different between groups.

All measured analytes (e.g., vitamins, minerals, trace elements) and plasma amino acids except for Phe remained within the normal range in both groups, although many patients in the group with irregular AAM intake had a protein intake and nutrient supply below the rec-

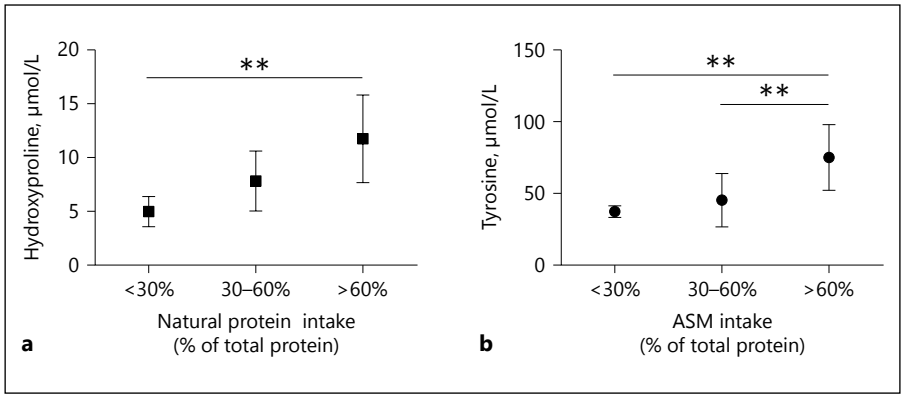

Fig. 3. Plasma hydroxyproline and natural protein intake. The concentration of hydroxyproline in plasma increases with the proportion of natural protein in the diet ($p = 0.005$ for trend; **a**). Plasma tyrosine increases with the amount of amino acid mixture consumed ($p = 0.004$ for trend; **b**). ** Significant differences between categories ($p < 0.05$ after Bonferroni correction).

ommended intake. However, some analytes in this group were significantly lower (e.g., vitamin B12, tyrosine), or showed a trend toward lower values (such as branched chain amino acids), although still remaining within the normal range. It is expected that these parameters may fall below the normal ranges in patients who consistently follow a rigid protein-restricted diet combined with very low AAM intake. Patients with irregular AAM intake participating in this study, however, largely continued AAM intake, albeit at lower than prescribed doses. Nonetheless, the observed trends indicate that these patients clearly are at risk for nutrient deficiencies [14, 15]. Therefore, regular follow-up with careful nutritional assessment by a specialised dietician is mandatory. Ordering of laboratory tests should be tailored according to the nutritional assessment, more extensive laboratory testing being justified in patients with poor treatment compliance (irregular AAM intake). Vitamin B12 deficiency may be the most frequent and relevant deficit in patients who relax their AAM intake [16].

Tyrosine as the product of the deficient enzyme phenylalanine hydroxylase becomes an essential amino acid in PKU [17]. For this reason, AAM are fortified with tyrosine. Accordingly, plasma tyrosine was significantly lower in patients with irregular AAM intake. Plasma tyrosine may, therefore, be used as a marker to estimate AAM intake.

Plasma hydroxyproline concentrations correlated with natural protein intake, as dietary hydroxyproline mainly derives from hydrolysed collagen products in animal protein (including gelatine products). Therefore, plasma hydroxyproline may represent a biomarker for estimating adherence to the protein-restricted diet, at least in patients who do not remain on a strict vegetarian (or vegan like) diet. Accordingly, plasma glutamine levels were higher in group B, consuming higher amounts of natural protein. Meat products and eggs are relevant sources of dietary glutamine [18]. Hydroxyproline also represents a bone resorption marker, which in theory may confound the association with natural protein intake. However, it is unlikely that bone resorption will be a major confounding factor in the present group of patients, as we would not expect relevant variations in bone resorption in adults with mature skeleton and premenopausal females. Other bone resorption markers such as carboxy-terminal collagen (CTX) crosslinks were not routinely measured. The value of hydroxyproline as a biomarker for natural protein intake will need to be validated in further studies.

This study represents the typical situation and real-life setting of an adult PKU clinic, where many patients who stay in regular follow-up principally remain on treatment, but relax their diet (restriction of natural protein, AAM intake) to a variable degree [8, 9]. Commonly, patients reduce the amount of AAM, but maintain some dietary protein restriction. This behavior puts patients at risk for nutritional deficiencies and imbalances. In this study, this is demonstrated for the group of patients with inadequate AAM intake. Although patients in this group are less adherent to treatment, they are not completely off-diet, and still achieve reasonable metabolic control in terms of Phe concentrations. However, even patients in the group with regular AAM intake on average have Phe values somewhat above the recommended target in Switzerland, which illustrates the common problem that even adults with good treatment adherence will often struggle to maintain a rigid protein-restricted diet to reach recommended Phe targets. Even in the group with good compliance (group A), many patients take the total AAM dose in 2 portions daily instead of 3 as recommended. Nonetheless, they achieved sufficient nutrient supply as long as the total daily amount of AAM was adequate. No differ-

ences in nutrient supply and the examined laboratory parameters were found in patients taking their AAM in 2 portions daily when compared to patients with 3 portions daily (data not shown).

Limitations of this study are the small group sizes, especially for the group with irregular AAM intake. Some of the trends observed may have reached significance with larger group sizes. Patients with better adherence to the treatment regimen in general were easier to motivate for study participation, which is a common problem in clinical studies.

In conclusion, this study demonstrates that patients who lower their AAM intake are at risk for nutritional deficiencies and have distinct dietary patterns compared to patients who follow the prescriptions more strictly. This underlines the importance of a regular medical and nutritional assessment, support, and long-term follow-up of adults with PKU in dedicated centers for metabolic disorders.

Acknowledgment

This work was in part supported by "radiz – Rare Disease Initiative Zurich, Clinical Research Priority Program University of Zurich." The authors thank those patients who participated in this study. We thank Fabian Meienberg and Stefanie Klein for dietetic advice and instruction of patients at the University Hospital Basel.

Disclosure Statement

S.B. was supported by a grant from Milupa Metabolics.

Author Contribution

M.H. and M.R.B. designed the research. M.H., S.B., C.T., and J.R. conducted the research. M.H., S.B., and C.T. analysed the data. M.H., S.B., C.T., P.G., and M.R.B. wrote or critically revised the manuscript. All authors have approved the final article.

References

1 Blau N, van Spronsen FJ, Levy HL: Phenylketonuria. Lancet 2010;376:1417–1427.
2 Donlon J, Sarkissian C, Levy H, Scriver CR: Hyperphenylalaninemia: phenylalanine hydroxylase deficiency; in Beaudet AL, Vogelstein B, Kinzler KW, Antonarakis SE, Ballabio A, Gibson KM, Mitchell G (eds): The Online Metabolic and Molecular Bases of Inherited Disease. New York, The McGraw-Hill Companies, Inc., 2014.
3 MacDonald A, Rocha JC, van Rijn M, Feillet F: Nutrition in phenylketonuria. Mol Genet Metab 2011;104(suppl):S10–S18.
4 Lammardo AM, Robert M, Rocha JC, van Rijn M, Ahring K, Belanger-Quintana A, MacDonald A, Dokoupil K, Ozel HG, Goyens P, Feillet F: Main issues in micronutrient supplementation in phenylketonuria. Mol Genet Metab 2013;110(suppl):S1–S5.
5 Vockley J, Andersson HC, Antshel KM, Braverman NE, Burton BK, Frazier DM, Mitchell J, Smith WE, Thompson BH, Berry SA; American College of Medical Genetics and Genomics Therapeutics Committee: Phenylalanine hydroxylase deficiency: diagnosis and management guideline. Genet Med 2014;16:188–200.
6 van Spronsen FJ, van Wegberg AM, Ahring K, Belanger-Quintana A, Blau N, Bosch AM, Burlina A, Campistol J, Feillet F, Gizewska M, Huijbregts SC, Kearney S, Leuzzi V, Maillot F, Muntau AC, Trefz FK, van Rijn M, Walter JH, MacDonald A: Key European guidelines for the diagnosis and management of patients with phenylketonuria. Lancet Diabetes Endocrinol 2017;pii:S2213-8587(16)30320-5.
7 Trefz FK, van Spronsen FJ, MacDonald A, Feillet F, Muntau AC, Belanger-Quintana A, Burlina A, Demirkol M, Giovannini M, Gasteyger C: Management of adult patients with phenylketonuria: survey results from 24 countries. Eur J Pediatr 2015;174:119–127.
8 Das AM, Goedecke K, Meyer U, Kanzelmeyer N, Koch S, Illsinger S, Lucke T, Hartmann H, Lange K, Lanfermann H, Hoy L, Ding XQ: Dietary habits and metabolic control in adolescents and young adults with phenylketonuria: self-imposed protein restriction may be harmful. JIMD Rep 2014;13:149–158.
9 Walter JH, White FJ, Hall SK, MacDonald A, Rylance G, Boneh A, Francis DE, Shortland GJ, Schmidt M, Vail A: How practical are recommendations for dietary control in phenylketonuria? Lancet 2002;360:55–57.
10 MacDonald A, Gokmen-Ozel H, van Rijn M, Burgard P: The reality of dietary compliance in the management of phenylketonuria. J Inherit Metab Dis 2010;33:665–670.
11 Evans S, Daly A, MacDonald J, Preece MA, Santra S, Vijay S, Chakrapani A, MacDonald A: The micronutrient status of patients with phenylketonuria on dietary treatment: an ongoing challenge. Ann Nutr Metab 2014;65:42–48.
12 Rohde C, von Teeffelen-Heithoff A, Thiele AG, Arelin M, Mutze U, Kiener C, Gerloff J, Baerwald C, Schultz S, Heller C, Muller AS, Kiess W, Beblo S: PKU patients on a relaxed diet may be at risk for micronutrient deficiencies. Eur J Clin Nutr 2014;68:119–124.
13 Deutsche Gesellschaft für Ernährung ÖGfE, Schweizerische Gesellschaft für Ernährungsforschung, Schweizerische Vereinigung für Ernährung (Hrsg.): Referenzwerte für die Nährstoffzufuhr, ed 2. Aufl., 1. Ausgabe 2015. Bonn, Neuer Umschau Buchverl, 2015.
14 Robert M, Rocha JC, van Rijn M, Ahring K, Belanger-Quintana A, MacDonald A, Dokoupil K, Gokmen Ozel H, Lammardo AM, Goyens P, Feillet F: Micronutrient status in phenylketonuria. Mol Genet Metab 2013;110(suppl):S6–S17.
15 Crujeiras V, Aldamiz-Echevarria L, Dalmau J, Vitoria I, Andrade F, Roca I, Leis R, Fernandez-Marmiesse A, Couce ML: Vitamin and mineral status in patients with hyperphenylalaninemia. Mol Genet Metab 2015;115:145–150.
16 Hvas AM, Nexo E, Nielsen JB: Vitamin B12 and vitamin B6 supplementation is needed among adults with phenylketonuria (PKU). J Inherit Metab Dis 2006;29:47–53.
17 Hanley WB, Lee AW, Hanley AJ, Lehotay DC, Austin VJ, Schoonheyt WE, Platt BA, Clarke JT: "Hypotyrosinemia" in phenylketonuria. Mol Genet Metab 2000;69:286–294.
18 Schulpis KH, Kalogerakou M, Gioni V, Papastamataki M, Papassotiriou I: Glutamine, ornithine, citrulline and arginine levels in children with phenylketonuria: the diet effect. Clin Biochem 2011;44:821–825.